



NTP
National Toxicology Program

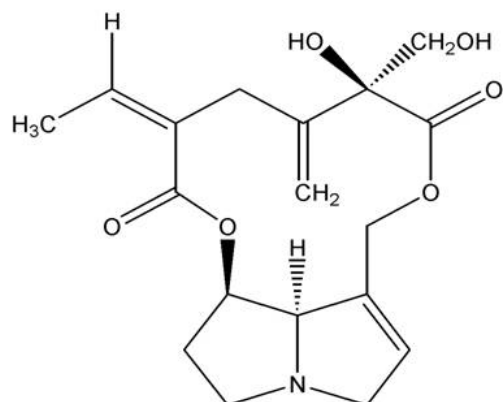
Report on Carcinogens Draft Substance Profile on Riddelliine

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Riddelliine



Riddelliine

13,19-didehydro-12,18-dihydroxysenecionan-11,16-dione
CAS No. 23246-96-0





Objectives

To present the science that supports the preliminary listing recommendation for riddelliine in the 12th RoC as *Reasonably Anticipated to be a Human Carcinogen*

- Information on use and exposure in US
- Cancer studies in humans and experimental animals
- Mechanistic evidence that supports the recommendation



Exposure



- Potential for significant US exposure
- Pyrrolizidine alkaloid (PA) produced by plants of *Senecio* genus (5 genera in US)
- Oral or dermal exposure
- Herbal products
- Contamination of food
 - Grains and flours
 - Honey
 - Milk (experimental)



Human Cancer Studies

- No studies identified on the relationship between human cancer and exposure to riddelliine



Sufficient Evidence from Studies in Experimental Animals

Early onset of tumors

NTP gavage studies in rats (F344/N) and mice (B6C3F₁)

13-wk exposure (both sexes)

Hepatocellular adenomas in female rats

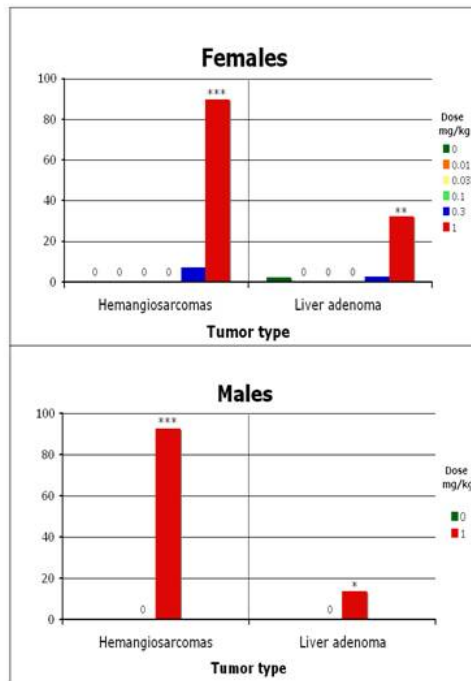
Tumors at multiple sites

NTP chronic gavage studies in rats (F344/N) and mice (B6C3F₁)

Two-year exposure (both sexes)

NTP conclusion: *clear evidence of carcinogenicity in rats and mice for both sexes*

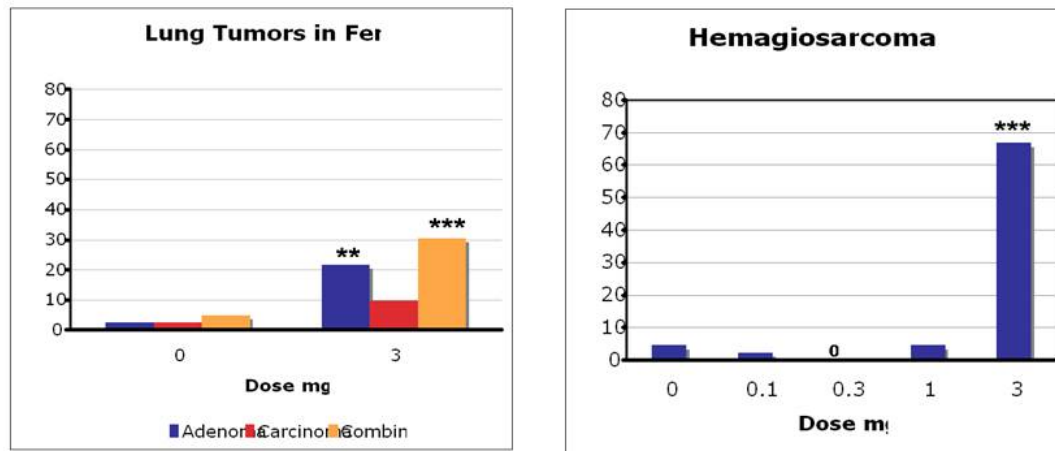
Liver Tumors in F344/N Rats Administered Riddelliine by Gavage for Two Years



- Very high incidence of hepatic hemangiosarcoma
- Increased incidence of liver tumors observed
- Increased incidence of mononuclear-cell leukemia in both sexes (data not shown)

* $P < 0.05$, ** < 0.01 , *** < 0.001 , survival-adjusted tumor incidence

Tumors in B6C3F₁ Mice Administered Riddelliine by Gavage for Two Years



- Increased incidence of lung tumors in female mice
- High incidence of hemangiosarcomas in male mice

** $P < 0.01$, *** $P < 0.001$, survival-adjusted tumor incidence

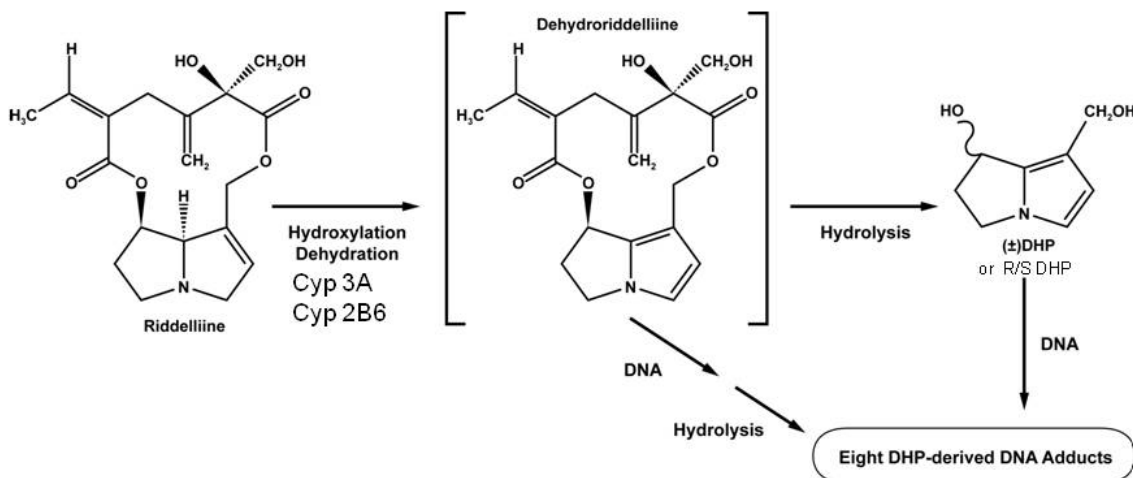


Mechanistic Evidence

- Proposed bioactivation pathways
- Genotoxicity
- Proposed mechanism of carcinogenicity in the liver



Hepatic Activation of Riddelliine Leading to DNA Adduct Formation





Carcinogenicity of Riddelliine Metabolites

- R-Dihydropyrrolizine (R-DHP)*
 - Dermal, female mice (malignant skin tumors)
 - Injection s.c., male mice (rhabdomyosarcomas)
- S-Dihydropyrrolizine (S-DHP)**
 - Injection i.p., male rats (tumors at multiple sites)

* Based on several studies

** Peterson *et al.* 1983



Genotoxicity

- *In Vitro*
 - Eight DHP-derived DNA adducts detected with riddelliine plus rat or human microsomes
 - Kinetic parameters of DHP formation similar for rat and human microsomes
 - Riddelliine, R-DHP, and S-DHP are mutagenic in bacteria
- *In Vivo*
 - Dose-dependent DHP adduct formation in livers of rats exposed to riddelliine

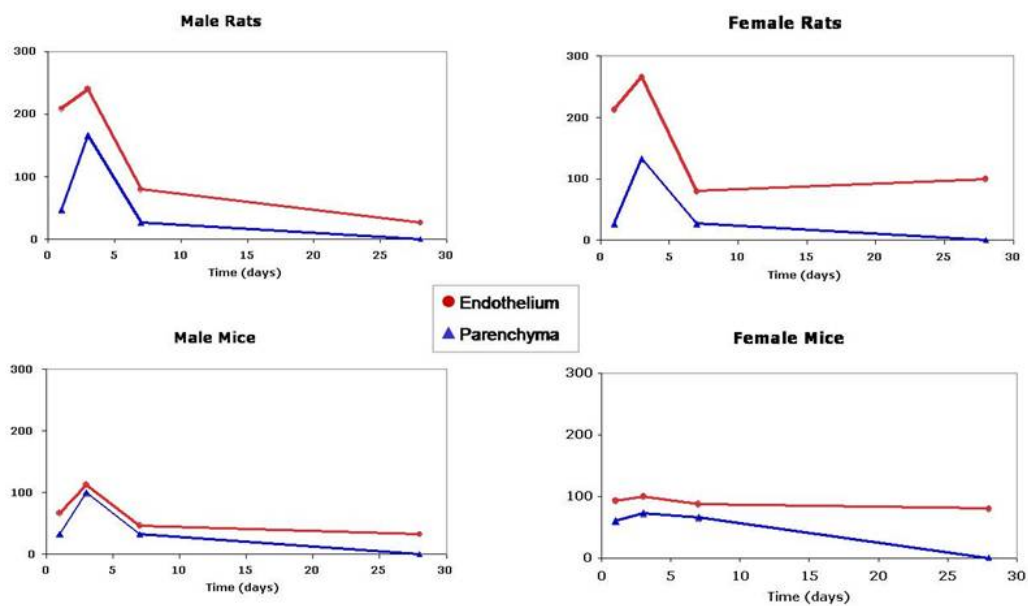


Proposed Mechanism of Carcinogenicity

- Hepatotoxicity in humans, livestock, and experimental animals from consumption of *Senecio* spp.
- Endothelial cell vs. hepatocyte
 - Higher and more persistent adduct levels in endothelial cells



DHP-derived DNA Adduct Levels in the Livers of F344 Rats and B6C3F₁ Mice



Chou *et al.* 2003



Proposed Mechanism of Carcinogenicity

- Hepatotoxicity in humans, livestock, and experimental animals from consumption of *Senecio* spp.
- Endothelial cell vs. hepatocyte
 - Higher and more persistent adduct levels in endothelial cells
 - Vascular endothelial growth factor (VEGF, endothelial cell mitogen) increased in hepatocytes
 - Higher frequency of mutations (GC to TA) in exposed endothelial cells
 - Mutations consistent with GC to TA transversions in *K-ras* in riddelliine-induced hemangiosarcomas



Proposed Riddelliine Listing

Riddelliine is *reasonably anticipated to be a human carcinogen* based on sufficient evidence of carcinogenicity from studies in experimental animals and supporting mechanistic data.